



## INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

<b>(51) International Patent Classification <sup>6</sup> :</b> <b>A61K 31/55, 31/70, 31/235, 31/515, 31/685</b>	<b>A1</b>	<b>(11) International Publication Number:</b> <b>WO 00/06174</b> <b>(43) International Publication Date:</b> 10 February 2000 (10.02.00)
<b>(21) International Application Number:</b> PCT/US99/17235 <b>(22) International Filing Date:</b> 30 July 1999 (30.07.99)  <b>(30) Priority Data:</b> 60/095,002                      31 July 1998 (31.07.98)                      US  <b>(71) Applicant:</b> MASSACHUSETTS INSTITUTE OF TECHNOLOGY [US/US]; 5 Cambridge Center, Room NE 25-230, Kendall Square, Cambridge, MA 02142-1324 (US).  <b>(72) Inventors:</b> WATKINS, Carol; Apartment 5-8C, 100 Memorial Drive, Cambridge, MA 02142 (US). WURTMAN, Richard, J.; Apartment 1205, 300 Boylston Street, Boston, MA 02116 (US).  <b>(74) Agents:</b> VILLACORTA, Gilberto, M. et al.; Pepper Hamilton LLP, 600 Fourteenth Street, N.W., Washington, DC 20005-2004 (US).		<b>(81) Designated States:</b> CA, JP, European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE).  <b>Published</b> <i>With international search report.</i>
<b>(54) Title:</b> METHODS FOR INCREASING CYTIDINE LEVELS IN VIVO AND TREATING CYTIDINE-DEPENDENT HUMAN DISEASES  <b>(57) Abstract</b>  Methods of treating certain neurological diseases using exogenous uridine or a uridine source alone as a precursor of endogenous cytidine, particularly in the human brain, are disclosed. Methods are also disclosed wherein exogenous uridine or a uridine source is combined either with drugs increasing uridine availability or with compounds that serve as a source of choline in phospholipid synthesis.		

**FOR THE PURPOSES OF INFORMATION ONLY**

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania	ES	Spain	LS	Lesotho	SI	Slovenia
AM	Armenia	FI	Finland	LT	Lithuania	SK	Slovakia
AT	Austria	FR	France	LU	Luxembourg	SN	Senegal
AU	Australia	GA	Gabon	LV	Latvia	SZ	Swaziland
AZ	Azerbaijan	GB	United Kingdom	MC	Monaco	TD	Chad
BA	Bosnia and Herzegovina	GE	Georgia	MD	Republic of Moldova	TG	Togo
BB	Barbados	GH	Ghana	MG	Madagascar	TJ	Tajikistan
BE	Belgium	GN	Guinea	MK	The former Yugoslav	TM	Turkmenistan
BF	Burkina Faso	GR	Greece		Republic of Macedonia	TR	Turkey
BG	Bulgaria	HU	Hungary	ML	Mali	TT	Trinidad and Tobago
BJ	Benin	IE	Ireland	MN	Mongolia	UA	Ukraine
BR	Brazil	IL	Israel	MR	Mauritania	UG	Uganda
BY	Belarus	IS	Iceland	MW	Malawi	US	United States of America
CA	Canada	IT	Italy	MX	Mexico	UZ	Uzbekistan
CF	Central African Republic	JP	Japan	NE	Niger	VN	Viet Nam
CG	Congo	KE	Kenya	NL	Netherlands	YU	Yugoslavia
CH	Switzerland	KG	Kyrgyzstan	NO	Norway	ZW	Zimbabwe
CI	Côte d'Ivoire	KP	Democratic People's	NZ	New Zealand		
CM	Cameroon		Republic of Korea	PL	Poland		
CN	China	KR	Republic of Korea	PT	Portugal		
CU	Cuba	KZ	Kazakhstan	RO	Romania		
CZ	Czech Republic	LC	Saint Lucia	RU	Russian Federation		
DE	Germany	LI	Liechtenstein	SD	Sudan		
DK	Denmark	LK	Sri Lanka	SE	Sweden		
EE	Estonia	LR	Liberia	SG	Singapore		

5       **METHODS FOR INCREASING CYTIDINE LEVELS IN VIVO AND TREATING**  
          **CYTIDINE-DEPENDENT HUMAN DISEASES**

BACKGROUND OF THE INVENTION

10      FIELD OF THE INVENTION

The present invention relates to methods of increasing cytidine levels by administering an exogenous uridine source and in particular to the pharmacological use of said uridine or uridine source alone or in combination with other pharmaceutical substances in treating certain neurological disorders.

15

DESCRIPTION OF THE RELATED ART

This invention stems from unexpected discovery that increase in levels of uridine following the administration of uridine or uridine source to certain animals comprising human patients, leads to increased levels of cytidine in a human body and particularly in the human brain. Thus, administering uridine or uridine precursors to human patients in need thereof can be as beneficial as administration of cytidine or cytidine precursors. However, the potential benefit of uridine or uridine source administration is overwhelmingly greater than the benefit of cytidine administration. This is due to the fact that cytidine, as opposed to uridine, either cannot cross or is much less efficient than uridine in crossing the blood-brain barrier (Cornford et al.,  
20      Independent blood-brain barrier transport systems for nucleic acid precursors. *Biochim. Biophys. Acta* 349:211-219, 1975).  
25

According to the knowledge relating to the metabolism of pyrimidine compounds, enzymes are known in the art, such as cytidine deaminase (EC 3.5.4.5), which converts cytidine into uridine. Cytidine deaminase can be found in some prokaryotes and eukaryotes including humans, primates, and some rodents although some species lack this enzyme. However, according to EC (enzyme classification) list there are no known examples of aminase-like enzymes, which are capable of opposite action, i.e., converting uridine into cytidine.  
30

The prior art relating to the process of uridine to cytidine conversion is also limited. Only one publication, citing two earlier references, seems to exist, wherein it was suggested that  
35      a soluble fraction of the rat liver and possibly of the brain may catalyze in vitro and in vivo the

5 conversion of uridine nucleotide to cytidine nucleotide (Dawson. Enzymic conversion of uridine nucleotide to cytidine precursor by rat brain. *J. Neurochem.* 15:31-34, 1968). Even though this report implicated the possibility of such an enzyme reaction in rats the activity of the enzyme does not appear to be sufficiently potent. As compared to the initial, administered dose of uridine (considered as 100%), the highest levels of newly converted cytidine in vivo  
10 were 12.4% in the liver and 9% in the brain. The conversion rates in vitro were 5.4% in the liver and 8.05% in the brain. Thus, maximum observed levels were within 5.4-12.4 % range. From a statistical point of view all these figures are within the range of a typical scatter in a gamma counter (15%) and practitioner in the art can dismiss them either as insignificant or irreproducible. Moreover, Dawson himself states that he was unable to recover a nucleotide  
15 with spectrophotometric characteristics of cytidine and admits that his conclusions were based on probabilistic guessing. Thus, the alleged phenomenon observed by Dawson may have been due to misinterpretation of an experimental artifact as it is now known that experimentally measurable cytidine can be easily confused with tyrosine, which is chemically unrelated amino acid compound (see Fig. 1).

20 Thus, even though an enzyme catalyzing the conversion of uridine to cytidine may exist in rats its activity is not sufficiently potent to raise the levels of cytidine to a level that can be measured and ascertained beyond any doubt. Thus, these levels may be not sufficient to warrant practical exploitation for clinical application. Indeed nowhere in Dawson publication is there a suggestion or an attempt to make a suggestion that the uridine to cytidine conversion process  
25 can be useful for any medical modality. In addition, as it is the case with many other enzymes and metabolic pathways, this particular enzyme may have been present in rats but not in humans. One skilled in related art knows that a discovery of a biological process in one species of an animal, e.g., rat, does not necessarily means that a similar process is present in another animal, e.g., man. Based on that one skilled in the art will be not sufficiently motivated to  
30 exploit this phenomenon for any useful purposes other than an experimental tool to study enzyme metabolism in rats. Consequently, the prior art is silent in regard to the use of the process of uridine to cytidine conversion for any meaningful application.

Uridine is a pyrimidine nucleoside and is essential in the synthesis of ribonucleic acids and tissue glycogens such as UDP glucose and UTP glucose. Medical uses of uridine alone are  
35 limited to treatment of genetic disorders related to deficiencies of pyrimidine synthesis such as

5     orotic aciduria (Becroft DM, et al., Hereditary orotic aciduria: long-term therapy with uridine and a trial of uracil. *J Pediatr.* 1969 Nov; 75(5): 885-891). Other less common uses of uridine alone are known such as treatment of seizures and epilepsy (Roberts CA, et al., Uridine anticonvulsant effects: selective control of nucleoside incorporation in experimental epilepsy. *Epilepsia.* 1974 Dec; 15(4): 479-500). Most commonly, uridine is used in combination with  
10     cytidine (Monticone GF, et al., On the therapeutic use of the nucleosides, cytidine and uridine, in some neurological diseases. *Minerva Med.* 1966 Dec 19; 57(101): 4348-4352). The uses of this particular dual combination range from liver and kidney diseases to a number of neurological and cerebrovascular diseases but such uses are irrelevant to the present invention directed at the use of uridine without concomitant use with cytidine.

15             U.S. Pat. No. 4,960,759, issued to De Luca et al., on October 2, 1990 discloses the pharmacological use of uridine in the treatment of nervous disorders such as schizophrenia and Parkinson's disease. De Luca et al., teach that the benefit of uridine is due to increase in cholecystokinin levels in the brain, which in turn improves dopamine functioning and results in  
20     therapeutic benefit. Said benefit is described as a reduction in symptoms of Parkinson's disease, which are tremor and rigidity. As the preferred embodiment of the instant invention is treatment of neurological disorders unrelated to schizophrenia and Parkinson's disease it is clear that the teachings by De Luca et al., are irrelevant to this invention.

           The U.S. Pat. No. 5,470,838, issued to von Borstel et al., on November 28, 1995 discloses the method of delivering exogenous uridine or cytidine in form of acylated uridine or  
25     cytidine and said compounds as useful in treating cardiac insufficiency, myocardial infarction, and cirrhosis of the liver. Von Borstel et al., propose to use both forms of pyrimidines since it was not obvious to them that uridine alone is effective. The absolute requirement of both cytidine and uridine was due to the lack of knowledge and anticipation in the prior art that uridine might convert into cytidine, especially in humans. One skilled in the art will recognize  
30     that the disclosed composition matter is different and diseases to be treated are not the same as in the present invention.

           The U.S. Pat. Nos. 5,141,943; 5,567,689; and 5,723,449 disclose various methods and compositions to raise levels of uridine in the blood as useful for reducing toxicity of pyrimidine nucleoside drugs such as AZT and 5-Fluouracil for AIDS and cancer therapy respectively. It is

5     apparent to anyone skilled in the art that these teachings have nothing in common with the present invention.

          Although all of these patents and prior art references disclose at least one or another aspect of the instant invention none of them taught specifically that cytidine levels can be raised in humans by administering uridine or uridine source as useful for the treatment of certain  
10    neurological or brain disorders. These disorders comprise disorders associated with aging such as memory decline and age related decline in cognition functions. These disorders also comprise memory decline and related cognition dysfunction associated with pathological conditions like Alzheimer's disease, Pick's disease, Lewy Body disease, and/or dementias like Huntington's disease and AIDS dementia. Other cognitive dysfunctions, i.e., disorders of attention, alertness,  
15    concentration, focus, and dyslexia can also be treated. Other uses of uridine therapy can be imagined such as treatment of mood and emotional disorders, e.g., mania, depression, stress, panic, anxiety, insomnia, dysthemia, psychosis, seasonal effective disorders and bipolar disorders. Neurological diseases like ataxias, including Friedreich's ataxia and movement disorders like tardive dyskinesia can also be treated. Method of treating stroke, cerebral  
20    thrombosis, ischemia, and related cerebrovascular diseases resulting from hypoxia as well as behavioral and neurological syndromes seen after brain trauma, spinal cord injury and/or anoxia can be also imagined. Methods of treating diseases of the peripheral nervous system, e.g., neuromuscular disorders like myasthenia gravis, the post-polio syndrome, and muscular dystrophies are also possible. It is also possible to imagine the methods of treating neurological  
25    diseases associated with dopaminergic pathway, e.g., schizophrenia and Parkinson's disease as treated by combination therapy in which uridine is one of constituents.

          Thus, none of the prior art patents or references have anticipated or made the instant invention obvious. The present invention is thus unique and stands out in the light of the prior art.

#### 30     SUMMARY OF THE INVENTION

          This invention is based upon unexpected discovery that uridine administration in humans leads to increase in systemic and brain cytidine. While certain methods of treatment of certain neurological diseases using uridine are known the prior art has heretofore failed to provide

5 methods of treatment of diseases which are objects of this invention. These and other objects of the invention will become more readily apparent from the following description.

It is a general object of this invention to provide a novel method of treatment using uridine, uridine prodrugs, and pharmaceutically derivatives thereof alone or in combination with compounds that are useful in increasing uridine availability. The terms "uridine precursor" or  
10 "uridine source" or "uridine prodrug" are used interchangeably and as defined hereinafter mean compounds, e.g., uridine salts or food products containing uridine, that transform into uridine upon administration to a host such as human.

The present invention also provides a method of treating various neurological disorders comprising memory disorders associated with aging as well as memory decline and cognitive  
15 dysfunction associated with pathological conditions like Alzheimer's disease, Pick's disease, Lewy Body disease, and/or dementias like Huntington's disease and AIDS dementia.

The present invention also provides a method of slowing down or reversing brain-aging process, which may cause memory decline and cognitive dysfunction.

Other uses of uridine therapy are provided such as treatment of cognitive dysfunction,  
20 i.e., disorders of attention, alertness, concentration, focus, and dyslexia.

Other uses of uridine therapy are also provided such as treatment of mood and emotional disorders, e.g., mania, depression, stress, panic, anxiety, insomnia, dysthemia, psychosis, seasonal effective disorders and bipolar disorders.

It is also an object of this invention to provide methods for treating other neurological  
25 diseases like ataxias, including Friedreich 's ataxia.

It is also an object of this invention to provide methods for treating strokes, cerebral thrombosis, ischemia, and related cerebrovascular diseases resulting from hypoxia.

Methods of treating behavioral and neurological syndromes seen after brain trauma, spinal cord injury and/or anoxia are also provided.

30 The present invention also provides a method of treating patients with movement disorders like tardive dyskinesia.

Methods of treating diseases of the peripheral nervous system, e.g., neuromuscular disorders like myasthenia gravis, the post-polio syndrome, and muscular dystrophies are also provided.

5 It is also an object of this invention to provide methods for treating other diseases known in the art and involving or dependent on cholinergic or uridine/cytidine metabolic pathways.

Methods of treating neurological diseases associated with dopaminergic pathway, e.g., schizophrenia and Parkinson's disease as treated by combination therapy in which uridine is one  
10 of constituents are provided.

In another aspect, the invention provides additional pharmaceutical compositions that can inhibit metabolic degradation or clearance of uridine as compounds enhancing uridine bioavailability. Among such compounds are uridine phosphorylase inhibitors like 5-benzyl barbiturate or derivatives thereof. Among such compounds are also compounds competing with  
15 uridine in renal transport mechanism. They comprise compounds like L-uridine, L-2',3'-dideoxyuridine, and D-2',3'-dideoxyuridine. Among such compounds are also uridine secretion inhibiting compounds like dilazep or hexobendine.

Choline is involved in metabolism and lipid transport and is a component of a number of important biological compounds including the membrane phospholipids like lecithin and  
20 sphingomyelin. Choline is also a precursor of acetylcholine - one of most important neurotransmitters. Although a required nutrient for several animal species, choline is not currently designated as essential for humans. However, recent clinical studies show it to be essential for normal liver function. Additionally, a large body of evidence from the fields of molecular and cell biology shows that certain phospholipids play a critical role in generating  
25 second messengers for cell membrane signal transduction. This process involves a cascade of reactions that translate an external cell stimulus such as a hormone or growth factor into a change in cell transport, metabolism, growth, function, or gene expression. Disruptions in phospholipid metabolism can interfere with this process and may underlie certain disease states such as cancer and Alzheimer's disease. However, choline alone is not useful as a therapeutical  
30 modality. In light of the instant invention choline or choline precursors are appropriate in considering them in combination with uridine or uridine source.

It is thus a further object of this invention to establish a synergy between uridine and various compounds affecting cholinergic pathway and/or phospholipid metabolism. Among them are CDP-choline, choline, choline salts, lecithin or phosphatidylcholine,



- 5 phosphatidylethanolamine, various fatty acids, e.g., linoleic acid, and other known in the art compounds or mixtures thereof involved in phospholipid synthesis.

#### BRIEF DESCRIPTION OF THE DRAWINGS

- 10 Fig. 1 illustrates the coincidence of cytidine and tyrosine peaks (6.59) when tested by a standard HPLC method.

Fig. 2 illustrates distinct cytidine (3.25) and tyrosine (2.92) peaks when tested by a modified HPLC method, which utilizes elution buffer with low methanol.

- Fig. 3 shows the ratio of uridine (100%) to cytidine in plasma after oral administration of 250  
15 milligram per kg of body weight (mg/kg) of uridine.

Fig. 4 shows the ratio of uridine (100%) to cytidine in the brain after oral administration of 250 milligram per kg of body weight (mg/kg) of uridine.

#### DESCRIPTION OF THE PREFERRED EMBODIMENTS

- 20 The present invention concerns methods and compositions for increasing systemic and brain levels of cytidine in a human patient via administration of uridine or uridine source alone. Also methods and compositions are contemplated which encompass the use of uridine or uridine source in combination with drugs that increase uridine availability. Among such drugs are drugs that act as uridine phosphorylase inhibitors like benzyl barbiturate or derivatives  
25 thereof. Among such drugs are also drugs that act as uridine secretion inhibiting compounds like dilazep or hexobendine. Among such drugs are drugs that act as uridine renal transport competitors like L-uridine, L-2',3'-dideoxyuridine, and D-2',3'-dideoxyuridine. Also methods and compositions are disclosed that are beneficial to a human patient in need thereof and which act in synergy with uridine in generation of phospholipids involved in brain cell membrane  
30 formation and repair. More specifically choline-based compounds are contemplated as compounds acting in synergy with uridine or uridine source. Among them are choline, choline salts or esters, such as choline bitartrate or stearate or the like, or compound that dissociate to choline, such as sphingomyelin, cytidine-diphospho-choline or citicoline or CDP-choline, acylglycerophosphocholines, e.g., lecithin, lysolecithin, glycerophosphatidylcholine, mixtures  
35 thereof or the like.

5           Neurological diseases associated with dopaminergic pathway, e.g., schizophrenia and Parkinson's disease as treated by uridine alone are specifically excluded.

          Other neurological conditions that were suggested to be treatable by uridine like epilepsy, seizures, and convulsions are excluded.

          Specific medical uses of uridine in combination with cytidine known in the prior art are  
10       also excluded.

          The following examples illustrate the invention and are not intended to be limiting unless otherwise specified.

#### EXAMPLE 1.

15           In this example a new method is established that overcomes the problem of the coincidence of cytidine and tyrosine peaks when tested by a standard HPLC method for measuring various nucleosides in biological fluids (see Fig. 1). By using the standard HPLC method, one can, however, easily distinguish the uridine peak from the cytidine peak. Detailed description of the HPLC method can be found for example in Lopez-Coviella et al., (Evidence  
20       that 5'-cytidinephosphocholine can affect brain phospholipid composition by increasing choline and cytidine plasma levels. *J. Neurochemistry* 65: 889-894, 1995). Modified HPLC is carried out in a same way as standard HPLC except that elution buffer contains low amount of methanol (0.1%) instead of formic acid and as a result cytidine can be distinguished from unrelated compound tyrosine (Fig. 2). This method is useful in distinguishing cytidine from  
25       masking effect of amino acid tyrosine, which may concomitantly be present in tested biological fluid, e.g., plasma or cerebrospinal fluid (CSF). Due to the overlap between cytidine and tyrosine it is very likely that the results of all the prior art studies dealing with cytidine measurement, including present inventors' own studies supra, were interpreted incorrectly.

#### 30       EXAMPLE 2.

          Gerbils rather than rats or other rodents are selected for this example, as the pyrimidine metabolism of said gerbils is closer to humans. For practical and ethical reasons humans cannot always be used for certain experimental studies and those skilled in the art generally recognize that the gerbil model is equivalent to a human model. Indeed, gerbils are the choice model for  
35       certain human diseases and brain disorders such as cerebral ischemia (Ginsburg et al., Rodent

5 models of cerebral ischemia. *Stroke* 20:1627-1642, 1989). Gerbils are given orally uridine and 60 minutes later plasma and brain levels of cytidine and uridine are measured by modified HPLC method described in Example 1. The Fig. 3 shows the relative ratio between uridine and cytidine levels in plasma after oral administration of 250 milligram per kg of body weight (mg/kg) of uridine. The Fig. 4 shows the relative ratio between uridine and cytidine levels in the  
10 brain after oral administration of 250 mg/kg of uridine. These results indicate that the metabolic processing of uridine in the brain is different than systemic processing of uridine in plasma. The results also indicate that uridine, when transported into the brain, is readily converted to cytidine and this conversion is more efficient in the brain than in plasma. Similar experiments are also carried out in humans wherein instead of measuring brain levels of nucleosides the CSF levels  
15 are measured. The finding that uridine is readily converted to cytidine especially in the brain is totally unexpected and constitutes the basis for the present invention.

### EXAMPLE 3

In Example 3 a clinical study is carried out with the goal of treating memory disorders  
20 and cognitive dysfunctions associated with aging as well as memory decline and cognitive dysfunction associated with pathological conditions like Alzheimer's disease, Pick's disease, Lewy Body disease, and/or dementias like Huntington's disease and AIDS dementia. Patients with non-pathological dementia associated with aging are also included. Oral doses of uridine alone ranging from 5 mg to 50,000 mg are administered daily to five male and five female  
25 patients suffering from one of the diseases listed above. The adjustment in dosage to select optimally effective pharmaceutical dose is a routine procedure well known to the practitioner skilled in the relevant art. The terms "therapeutically" or "pharmaceutically" or "pharmacologically effective dose" of a drug as used hereinafter means the amount (dosage) of the drug that provides desired clinical effect in at least 10% of treated patients" population.

30 Various other uridine-based compounds other than uridine itself serve as uridine source or uridine precursors. These are uridine-rich food or dietary products like algae; salts of uridine like uridine phosphates, acylated uridine or the like. These also include compounds, like CDP-choline, though structurally unrelated to uridine are able, nevertheless, to raise uridine levels in treated subjects. If required by the exigency of the therapy, therapeutically or pharmacologically  
35 effective doses of CDP-choline are also administered since it is now known that administration

5 of said drug raises uridine but not cytidine levels and as such CDP-choline or citicoline is by definition the uridine source.

If required by the exigency of the therapy, therapeutically or pharmacologically effective doses of acyl derivatives of uridine or mixtures thereof like those disclosed in U.S. Pat. No. 5,470,838 are also administered.

10 If required by the exigency of the therapy, therapeutically or pharmacologically effective doses of uridine phosphorylase inhibitors like 5-benzyl barbiturate derivatives or mixtures thereof as disclosed in U.S. Pat. No. 5,141,943 are also administered.

If required by the exigency of the therapy, therapeutically or pharmacologically effective doses of uridine secretion inhibiting compounds like dilazep, hexobendine, or mixtures thereof as disclosed in U.S. PAT. No. 5,567,689 are also administered.

If required by the exigency of the therapy, therapeutically or pharmacologically effective doses of compounds which compete with uridine in kidney clearance like L-uridine, L-2',3'-dideoxyuridine, and D-2',3'-dideoxyuridine or mixtures thereof as disclosed in U.S. Pat. Nos. 5,723,449 and 5,567,689 are also administered. Therapeutically or pharmacologically effective doses of uridine as defined herein are also doses that produce blood or brain levels of cytidine ranging between 0.1 micromole ( $\mu\text{M}$ ) and 1 millimole ( $\text{mM}$ ). In general, therapeutically or pharmacologically effective doses as defined herein are also doses of drugs combination, which produce desired effect in at least 10% of treated patients= population. Doses are administered either as a single dose or divided in several doses. Drugs are administered orally such as in tablet, capsule or liquid form or parenterally by intravenous, intramuscular or subcutaneous injection.

When necessary and as required by the exigency of the therapy uridine is administered in combination with other compounds that act either synergistically or in additive manner. This reduces the therapeutic dose of administered drugs, thereby reducing potential undesirable side effects and frequency of drug administration. Compounds that act in such a manner are chemical substances participating in cholinergic metabolism. For example, compounds administered along with uridine are following choline-based compounds: choline, choline salts or esters, such as choline bitartrate or stearate or the like, or compound that dissociate to choline, such as sphingomyelin, cytidine-diphospho-choline or citicoline or CDP-choline, acylglycerophosphocholines, e.g., lecithin, lysolecithin, glycerophosphatidylcholine, mixtures

5 thereof or the like. The choline or compound that dissociates into choline is administered so that a choline level of at least about 20-30 nanomoles and usually between 10 and 50 nanomoles is attained in patient's blood or brain.

The pharmacologically effective doses are within about 20 mg and 50 g/day range, preferably between about 100 mg and 10 g/day. Doses are administered either as a single dose  
10 or divided in several doses, e.g., 10 mg to 1 g/ cap or tab. The minimal duration of the therapy is at least one day but longer periods of time are usually required according to the exigency of the therapy. If needed, the usual time period spans from one day to the period of lifetime. When these compounds are not available in pure form the active ingredient comprises at least about 20-30 percent of the weight of the preparation. The clinical study is continued for at least 1 day  
15 or longer as required by the exigencies of the therapy. In general, the dose administered, the frequency of administration and the duration of the treatment will vary as a function of the condition of the patient and is determined according to standard clinical procedures known to the practitioner skilled in the relevant art.

#### 20 EXAMPLES 4-12

In Example 4 a clinical study is carried out, which by its design and principles is similar to clinical study of Example 3 except that patients enrolled in this study are patients with cognitive dysfunction, i.e., disorders of attention, alertness, concentration, focus, and dyslexia.

In Example 5 a clinical study is carried out, which by its design and principles is similar  
25 to clinical study of Example 3 except that patients enrolled in this study are patients with mood and emotional disorders, e.g., mania, depression, stress, panic, anxiety, insomnia, dysthemia, psychosis, seasonal effective disorders and bipolar disorders.

In Example 6 a clinical study is carried out, which by its design and principles is similar to clinical study of Example 3 except that patients enrolled in this study are patients with  
30 neurological diseases like ataxias, including Friedreich's ataxia.

In Example 7 a clinical study is carried out, which by its design and principles is similar to clinical study of Example 3 except that patients enrolled in this study are patients with movement disorders like tardive dyskinesia.

In Example 8 a clinical study is carried out, which by its design and principles is similar  
35 to clinical study of Example 3 except that patients enrolled in this study are patients with

5 strokes, cerebral thrombosis, ischemia, and related cerebrovascular diseases resulting from hypoxia.

In Example 9 a clinical study is carried out, which by its design and principles is similar to clinical study of Example 3 except that patients enrolled in this study are patients with behavioral and neurological syndromes seen after brain trauma, spinal cord injury and/or  
10 anoxia.

In Example 10 a clinical study is carried out, which by its design and principles is similar to clinical study of Example 3 except that patients enrolled in this study are patients with diseases of the peripheral nervous system, e.g., neuromuscular disorders like myasthenia gravis, the post-polio syndrome, and muscular dystrophies.

15 In Example 11 a clinical study is carried out, which by its design and principles is similar to clinical study of Example 3 except that patients enrolled in this study are patients with neurological diseases associated with dopaminergic pathway, e.g., schizophrenia and Parkinson's disease and said diseases are treated by combination therapy in which uridine is one of constituents.

20 In Example 12 a clinical study is carried out, which by its design and principles is similar to clinical study of Example 3 except that patients enrolled in this study are patients with other diseases known in the art and involving or dependent on cholinergic or uridine/cytidine metabolic pathways.

When possible clinical studies as disclosed in any preceding examples are preceded by  
25 in vivo studies in animal models, e.g., gerbil model, according to procedures established in the art.

While methods of treatment and pharmaceutical compositions recited in one of above examples have been described in detail, it will be apparent to those skilled in the art that these embodiments are exemplary rather than limiting and the true scope of the invention is that  
30 defined within the attached claims.

## 5 CLAIMS

What is claimed is:

- 10 1. A method of treating a human with neurological disorders comprising administering therapeutically effective doses of uridine or a uridine source.
2. The method according to claim 1 wherein said therapeutically effective dose of uridine or a uridine source is administered for at least one day.
3. The method according to claim 1 wherein said neurological disorders are memory disorders.
- 15 4. The method according to claim 3 wherein said memory disorders comprise memory decline associated with brain aging.
5. The method according to claim 3 wherein said memory disorders are selected from the group comprising of Pick's disease, Lewy Body disease, and/or dementias like Huntington's disease and AIDS dementia.
- 20 6. The method according to claim 1 wherein said neurological disorders are cognitive dysfunctions comprising lack of attention, alertness, concentration, focus, or dyslexia.
7. The method according to claim 1 wherein said neurological disorders are emotional disorders comprising mania, depression, stress, panic, anxiety, dysthemia, psychosis, seasonal effective disorders and bipolar disorders.
- 25 8. The method according to claim 1 wherein said neurological disorders are selected from the group consisting of ataxia or Friedreich's ataxia.
9. The method according to claim 1 wherein said neurological disorders are movement disorders comprising tardive dyskinesia.
10. The method according to claim 1 wherein said neurological disorders are cerebral thrombosis, ischemia, and related cerebrovascular diseases resulting from hypoxia.
- 30 11. The method according to claim 1 wherein said neurological disorders are behavioral and neurological syndromes.
12. The method according to claim 11 wherein said behavioral and neurological syndromes comprise those seen after brain trauma, spinal cord injury and/or anoxia.

- 5 13. The method according to claim 1 wherein said neurological disorders are peripheral nervous system disorders comprising neuromuscular disorders like myasthenia gravis, the post-polio syndrome, and muscular dystrophies.
14. The method according to claim 1 wherein said uridine is combined with pharmaceutically acceptable source of uridine.
- 10 15. The method according to claim 1 wherein said uridine or pharmaceutically acceptable source of uridine is administered in dosages of between 100 mg and 20 grams per day.
16. A method of treating a human with neurological disorders comprising administering therapeutically effective dose of at least uridine or a uridine source and other compounds.
17. The method according to claim 16 wherein said neurological disorders are memory disorders.
- 15 18. The method according to claim 17 wherein said memory disorders comprise memory decline associated with brain aging.
19. The method according to claim 17 wherein said memory disorders are selected from the group comprising Pick's disease, Lewy Body disease, and/or dementias like Huntington's disease and AIDS dementia.
- 20 20. The method according to claim 16 wherein said neurological disorders are cognitive dysfunctions comprising lack of attention, alertness, concentration, focus, or dyslexia.
21. The method according to claim 16 wherein said neurological disorders are emotional disorders comprising mania, depression, stress, panic, anxiety, dysthemia, psychosis, seasonal effective disorders and bipolar disorders.
- 25 22. The method according to claim 16 wherein said neurological disorders are selected from the group consisting of ataxia or Friedreich's ataxia.
23. The method according to claim 16 wherein said neurological disorders are movement disorders comprising tardive dyskinesia.
- 30 24. The method according to claim 16 wherein said neurological disorders are cerebral thrombosis, ischemia, and related cerebrovascular diseases resulting from hypoxia.
25. The method according to claim 16 wherein said neurological disorders are behavioral and neurological syndromes.
26. The method according to claim 25 wherein said behavioral and neurological syndromes comprise those seen after brain trauma, spinal cord injury and/or anoxia.
- 35



- 5 27. The method according to claim 16 wherein said neurological disorders are peripheral nervous system disorders comprising neuromuscular disorders like myasthenia gravis, the post-polio syndrome, and muscular dystrophies.
28. The method according to claim 16 wherein said neurological disorders selected from the group consisting of schizophrenia and Parkinson's disease.
- 10 29. The method according to claim 16 wherein said other compound is choline.
30. The method according to claim 16 wherein said other compounds are choline salt or stearate selected from the group consisting of choline chloride, choline bitartrate, choline stearate or mixtures thereof.
- 15 31. The method according to claim 16 wherein said other compounds are choline precursors selected from the group consisting of sphingomyelin, cytidine-diphospho-choline or citicoline or CDP-choline, an acylglycerophosphocholine, e.g., lecithin, lysolecithin, glycerophosphatidylcholine, fatty acids, or mixtures thereof.
32. The method as in one of claims 29-31 wherein a choline level of at least about 20-30 nanomoles and usually between 10 and 50 nanomoles is attained in patient=s blood or brain.
- 20 33. The method according to claim 16 wherein said other compounds are uridine phosphorylase inhibitors comprising benzyl barbiturate or derivatives thereof.
34. The method according to claim 16 wherein said other compounds are uridine secretion inhibiting compounds comprising dilazep or hexobendine.
35. The method according to claim 16 wherein said other compounds are uridine renal transport competitors selected from the group consisting of L-uridine, L-2',3'-dideoxyuridine, and D-25 2',3'-dideoxyuridine.
36. The method according to claim 16 wherein said other compound is a mixture of compounds from any one of claims 29-31 or 33-35.
37. The method according to claim 16 wherein said uridine or a uridine source are administered 30 in dosages of between 10 mg and 10 grams per day.
38. The method according to claim 16 wherein said therapeutically effective dose of uridine or a uridine source in combination with other drugs is administered for at least one day.

1/3

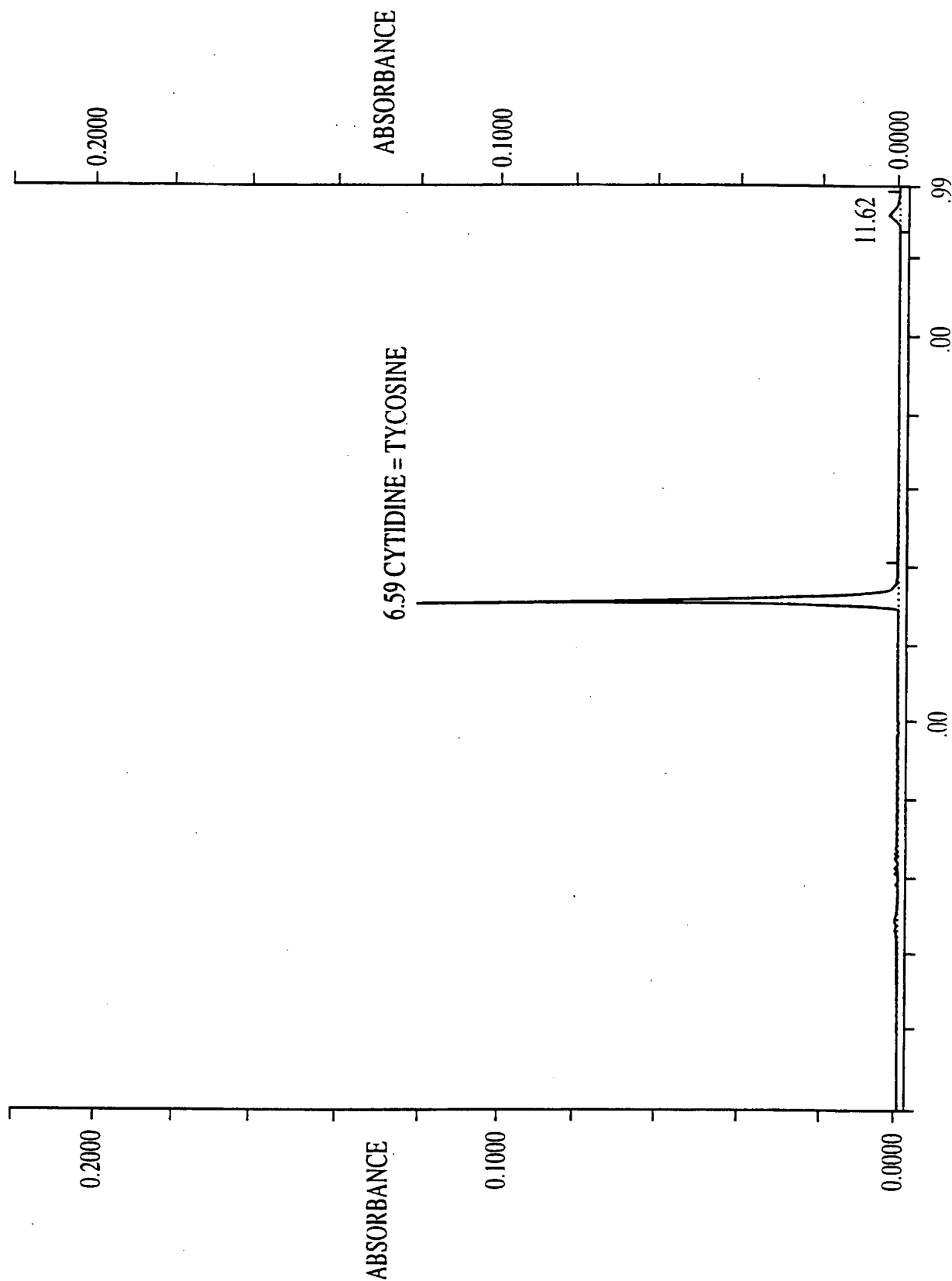


FIG. 1

2/3

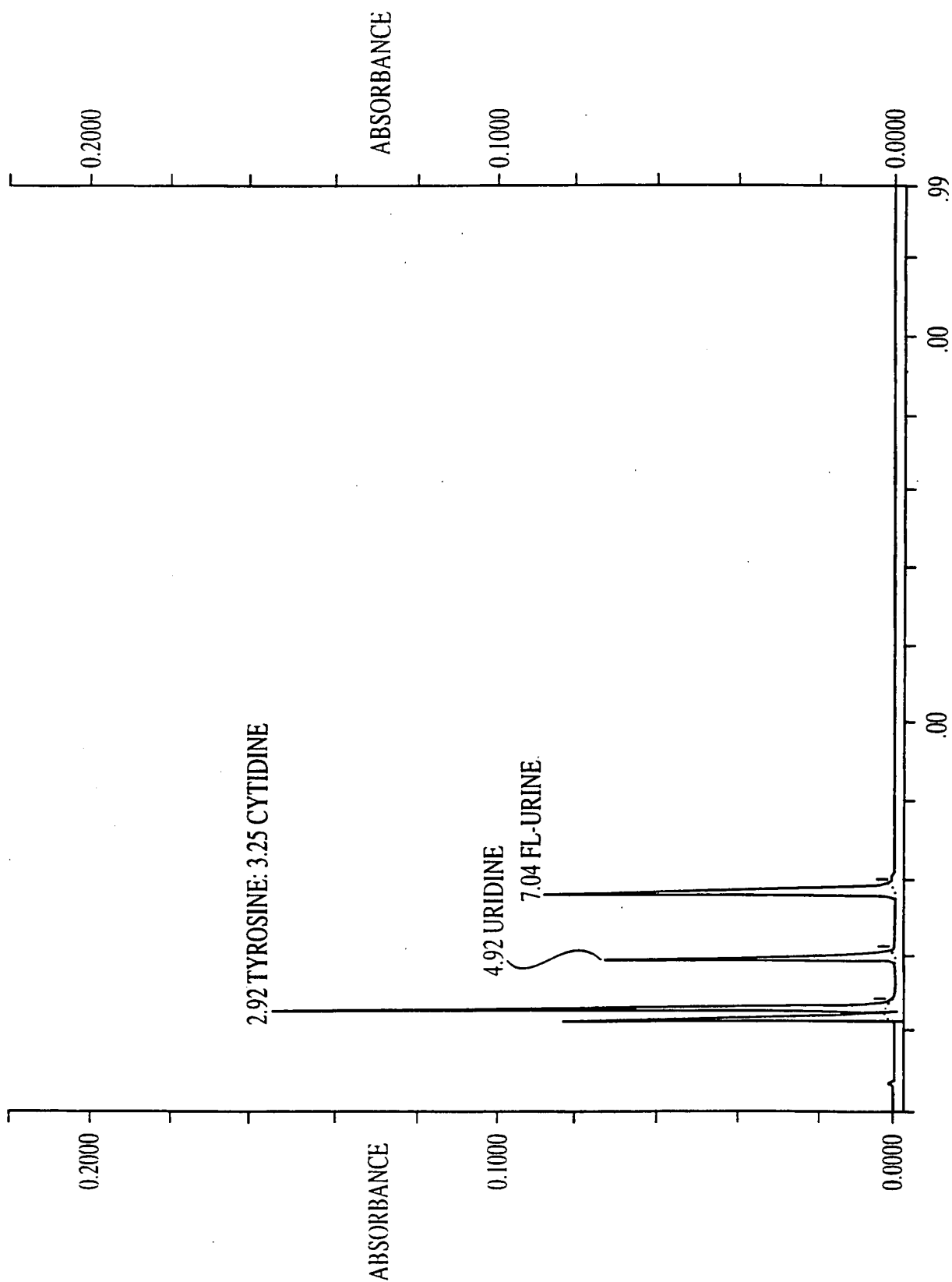


FIG. 2

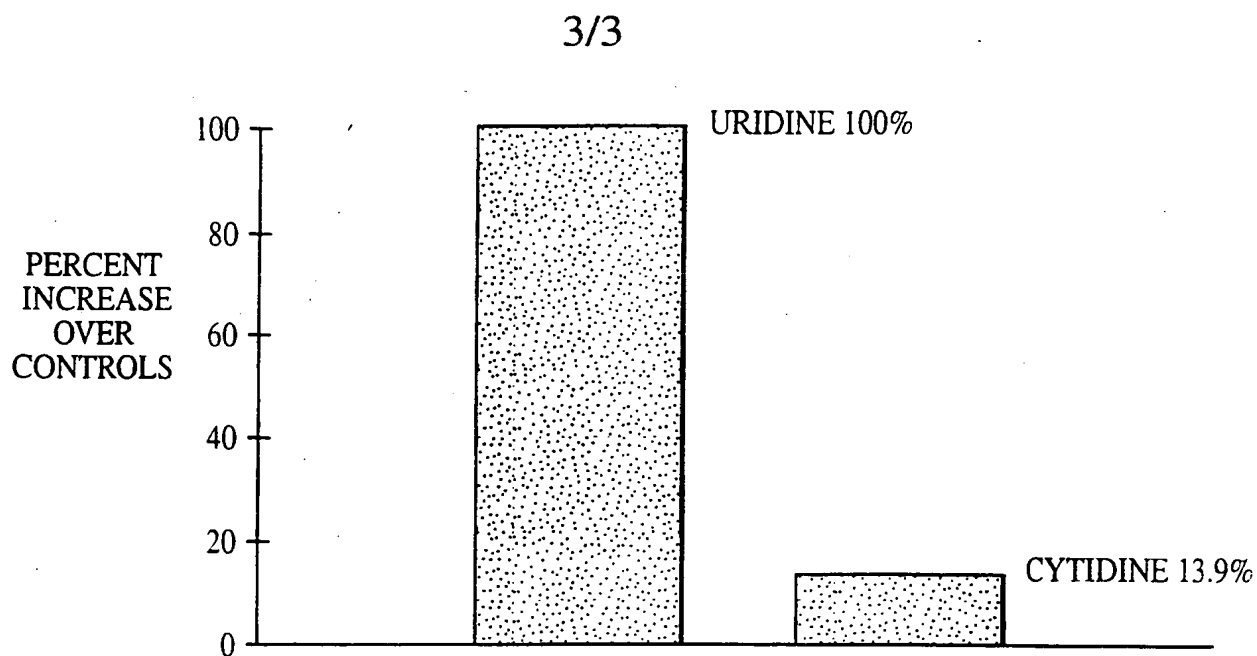


FIG. 3

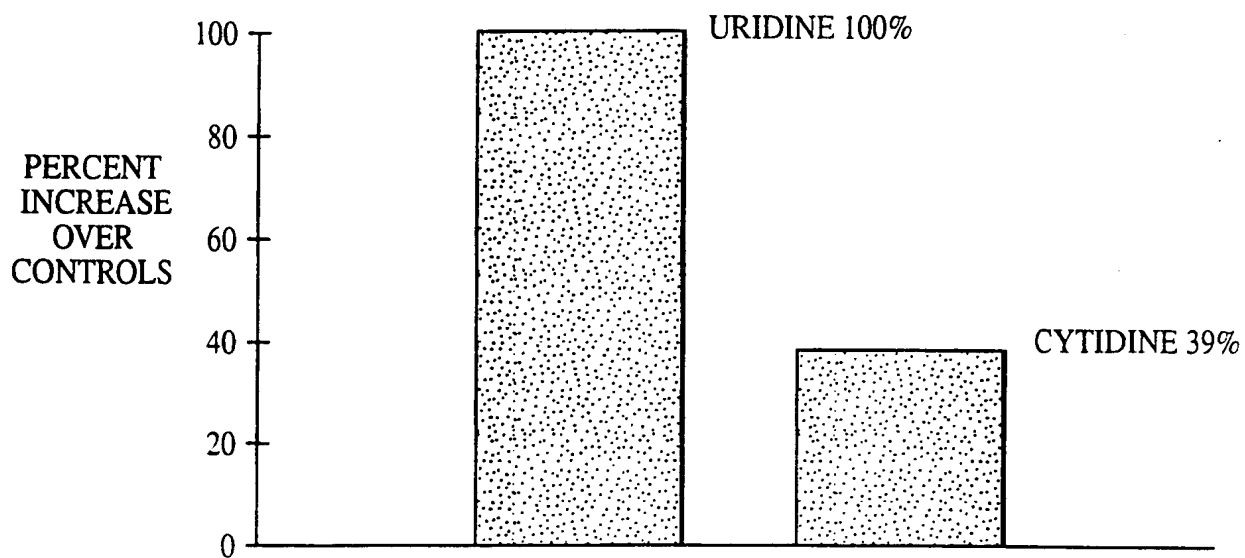


FIG. 4

## INTERNATIONAL SEARCH REPORT

International application No.  
PCT/US99/17235**A. CLASSIFICATION OF SUBJECT MATTER**

IPC(6) : A61K 31/55, 31/70, 31/235, 31/515, 31/685

US CL : 514/49, 50, 78, 218, 270, 533

According to International Patent Classification (IPC) or to both national classification and IPC

**B. FIELDS SEARCHED**

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 514/49, 50, 78, 218, 270, 533

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

REGISTRY, CA, CAPLUS, CAOLD, MEDLINE, EMBASE, WPIDS, BIOSIS

**C. DOCUMENTS CONSIDERED TO BE RELEVANT**

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 4,960,759 A (DE LUCA et al.) 02 October 1990), abstract.	1-38
X	US 5,567,689 A (SOMMADOSSI et al. ) 22 October 1996, column 1.	1-38
X	US 5,583,117 A (VON BORSTEL et al.) 10 December 1996, columns 6-7.	1-38
X	Database WPIDS on STN (Columbus, OH, USA), AN 1989-150746, WO 8903837 A, BAMAT, M.K. et al. New uridine- and cytidine acyl derivs - used in treatment e.g. of hepatopathy, diabetes, heart disease, cerebrovascular disorders and parkinson's disease. Abstract.	1-38

☒ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

* Special categories of cited documents:	*T* later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
*A* document defining the general state of the art which is not considered to be of particular relevance	*X* document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
*E* earlier document published on or after the international filing date	*Y* document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
*L* document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)	*&* document member of the same patent family
*O* document referring to an oral disclosure, use, exhibition or other means	
*P* document published prior to the international filing date but later than the priority date claimed	

Date of the actual completion of the international search

14 OCTOBER 1999

Date of mailing of the international search report

03 NOV 1999

Name and mailing address of the ISA/US  
Commissioner of Patents and Trademarks  
Box PCT  
Washington, D.C. 20231

Facsimile No. (703) 305-3230

Authorized officer

Rebecca Cook

Telephone No. (703) 308-1235

# INTERNATIONAL SEARCH REPORT

International application No.  
PCT/US99/17235

## C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	Database BIOSIS on STN (Columbus, OH, USA), AN 1996:466219, GALLAI, V. et al. 'Multi-infarct dementia: Modification of the P300 cognitive event-related potential in patients treated with the association of cytidine and uridine,' abstract, Ribista di Neuropsichiatria e Scienze Affini, 1995.	1-38
Y	Database BIOSIS on STN, (Columbus, OH, USA), AN 1996:26741, MYERS, C. et al. 'Uridine reduces rotation induced by L-dopa and methamphetamine in 6-OHDA-treated rats,' abstract, Pharmacology biochemistry and Behavior, 1995.	1-38
Y	Database CAPLUS on STN, (Columbus, OH, USA), No. 127:344939, PAGE, T. et al. 'Developmental disorder associated with increased cellular nucleotidase activity,' abstract, Proc. Natl. Acad. Sci. U.S.A., 1997.	1-38
X	Database CAPLUS on STN, (Columbus, OH, USA), No. 128:30410, WO 9745127 A1, PIAZZA, C. et al. Uridine-comprising therapeutic active agent for treatment of neurodegenerative disorders. Abstract.	1-38
X	Database CAPLUS on STN, (Columbus, OH, USA), No. 126:229629, JP 09030976 A2, YAMAMOTO, S., Purine nucleosides and pyrimidine nucleosides for the treatment of memory loss. Abstract.	1-38